For Research Use Only.
Not for use in diagnostic procedures .

Anti-cleaved IL-1β (Mouse) pAb

CODE No. PD039

CLONALITY Polyclonal
ISOTYPE Rabbit Ig, affinity purified
QUANTITY 100 µL

SOURCE Purified Ig from rabbit serum
REACTIVITY This antibody reacts with cleaved mouse IL-1β.
FORMULATION PBS containing 50% Glycerol (pH 7.2). No preservative is contained.
STORAGE This antibody solution is stable for one year from the date of purchase when stored at -20°C.

APPLICATION-CONFIRMED
Western blotting 1:500

SPECIES CROSS REACTIVITY on WB

Species	Human	Mouse	Rat	Hamster
Cell	Not tested	Mouse bone marrow derived macrophage stimulated with LPS + ATP or LPS + Salmonella infection	Not tested	Not tested
Reactivity		+		

Entrez Gene ID 16176 (Mouse)

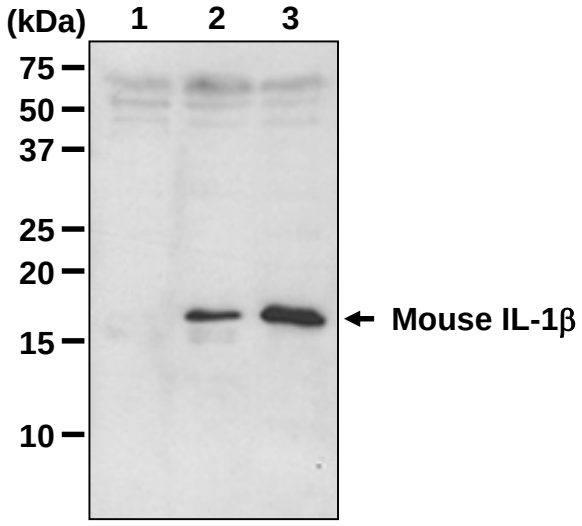
REFERENCES
1) Elinav, E., *et al.*, *Immunity* **34**, 665-679 (2011)
2) Schroder, K. and Tschopp, J., *Cell* **140**, 821-832 (2010)

For more information, please visit our web site **https://ruo.mbl.co.jp/.**

SDS-PAGE & Western blotting

1) Wash cells with PBS and suspend them in 1 mL of Laemmli's sample buffer, then sonicate briefly (up to 10 sec.).
2) Boil the samples for 3 min. and centrifuge. Load 10 μL of the sample per lane in a 1-mm-thick SDS-polyacrylamide gel (15% acrylamide) for electrophoresis.
3) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm^2 for 1 hr. in a semi-dry transfer system (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% methanol). See the manufacturer's manual for precise transfer procedure.
4) To reduce nonspecific binding, soak the membrane in 10% skimmed milk (in PBS, pH 7.2) for 1 hr. at room temperature.
5) Incubate the membrane with primary antibody diluted with Can Get Signal-1 (TOYOBO; code no. NKB-201) as suggested in the **APPLICATIONS** for 2 hr. at room temperature. (The concentration of antibody will depend on the conditions.)
6) Wash the membrane with PBS-T [0.05% Tween-20 in PBS] (5 min. x 3).
7) Incubate the membrane with the 1:5,000 Anti-IgG (Rabbit) pAb-HRP (MBL, code no. 458) diluted with 1% skimmed milk (in PBS, pH 7.2) for 1 hr. at room temperature.
8) Wash the membrane with PBS-T (5 min. x 3).
9) Wipe excess buffer on the membrane, and then incubate it with appropriate chemiluminescence reagent for 1 min. Remove extra reagent from the membrane by dabbing with paper towel, and seal it in plastic wrap.
10) Expose to an X-ray film in a dark room for 3 min. Develop the film as usual. The condition for exposure and development may vary.

(Positive controls for Western blotting; Mouse bone marrow derived macrophage stimulated with LPS + ATP and LPS + Salmonella infection)



Western blotting analysis of cleaved mouse IL-1β

Sample: Mouse bone marrow-derived macrophage
Lane 1: Stimulation with LPS for 3 hr.
Lane 2: Stimulation with LPS for 3 hr. and 5 mM ATP for 1 hr.
Lane 3: Stimulation with LPS for 3 hr. and infection with Salmonella for 1 hr.

Immunoblotted with Anti-cleaved IL-1β (Mouse) pAb (MBL, code no. PD039)